

Final report to:



Rufford Continuation Grant 2011

Conservation of killer whales (*Orcinus orca*) in the Russian Far East: promoting evidence of two separate species

Olga A. Filatova

MOSCOW STATE UNIVERSITY
named after M.V.Lomonosov



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Abstract

Killer whales are treated by Russian legislation as a “marine biological resource”; the species is not protected from environmental threats and quotas for live-capturing are granted annually. Although our previous work (funded by RSG) led to the cessation of killer whale captures in our project area of Eastern Kamchatka, the captors moved to other regions. In 2012, a young transient killer whale was captured in the western Okhotsk Sea. A key way to achieve better conservation for killer whales is to change their status in Russian legislation. Through this project we justified the recognition of two different killer whale species in Russian legislation. We demonstrated clear differences between resident and transient killer whales in the Russian Far East: in feeding ecology, in social structure, in acoustic behavior, in phenotypic features and, most importantly, in genetics. To disseminate the results among the scientists and the public, we gave many talks at conferences, workshops and meetings, wrote papers and letters to the officials.

Project leader: Olga A. Filatova

Address: Department of Vertebrate Zoology, Faculty of Biology, Moscow State University, Vorobiovy gory, 1/12, Moscow 119992, Russia

telephone +7-903-236-64-74

e-mail alazor@rambler.ru

website www.russianorca.com,

Conservation Issue: Main objective of our project is to justify the recognition of two different killer whale species in Russian legislation. My colleagues and I have studied orcas in Kamchatka since 2000. Our work funded by Rufford Small Grants helped determine killer whale status, stop live-captures for oceanariums in our study area and showed that commercial fisheries had a substantial negative impact on killer whale conservation. However, killer whales are still treated by Russian legislation as a “marine biological resource”; the species is not protected from environmental threats and quotas for live-capturing are still granted annually. Although our work led to the cessation of killer whale captures in our project area of Eastern Kamchatka, the captors moved to other regions. In 2012, a young transient killer whale was captured in the western Okhotsk Sea. A key way to achieve better conservation for killer whales on all aspects is to change their status in Russian legislation. Through this project we justified the recognition of two different killer whale species in Russian legislation. We demonstrated clear differences between resident and transient killer whales in the Russian Far East: in feeding ecology, in social structure,

in acoustic behavior, in phenotypic features and, most importantly, in genetics. Acceptance of two killer whale species in Russian legislation will lead to significant positive changes in their status and contribute to Russian whale conservation. Currently the number of killer whales in the Okhotsk Sea is estimated by Russian Federal Research Institute of Fisheries and Oceanography as 2,500, which is highly exaggerated. These numbers are considered to be high enough to support annual captures of 10 animals. The division of killer whales into two separate species will lead to re-estimation of their numbers. Since resident and transient killer whales are quite similar in appearance, only an experienced observer can tell the difference between them; therefore, the studies of species abundance will require the presence of killer whale specialists. This will represent the fundamental difference from the previous species abundance estimations, which were made by non-specialists with no reasonable basis and extrapolated without considering the species biology. Therefore, the new estimates are likely to give more realistic numbers totalling several hundred animals for each species, which should be too low to support captures.

Objectives:

- Analyse resident and transient killer whale feeding ecology, social structure, acoustic repertoires, genetic differentiation.
- Justify the existence of two separate killer whale species in Russian waters.
- Disseminate the results among the scientists and the officials.
- Disseminate the results among the public.

Results:

Field work

Our study took place in June-September 2011 at the Commander Islands (Russian Far East). We went to the sea in the inflatable boat with outboard motor and searched for killer whale groups to make photos and observe their behaviour. After killer whales were encountered the boat approached different groups of killer whales, followed each group in a distance of 20-50 m for about 10 – 20 min to take picture of each whale in the encounter. The photographs of the left side of individual whales were taken to show the details of dorsal fin and saddle patch, which allowed to distinguish individual whales using the technique of photoidentification. The data recorded during the work with a group included the date, time, duration of the work with group, location of the group, number of animals in the group, group composition and type of activity for the group. During feeding, we tried to

determine the prey items of killer whales. After taking photos, we made acoustic recordings of identified groups. When possible, we collected biopsy samples for genetic and stable isotope analysis.

Additional data

We have also obtained some data through collaboration with our colleagues working with killer whales in Southeastern Kamchatka (T.Ivkovich) and Western Okhotsk Sea (O.Shpak). This additional data allowed us to cover larger area and more populations by our analysis.

Lab analysis

First, we performed the analysis the phenotypic features based on obtained photographs of killer whales. We divided saddle patches into six types, including 1 to 2 open saddle patches, 3 to 5 semi-open saddle patches and 6 – closed (round) saddle patches (fig.1).

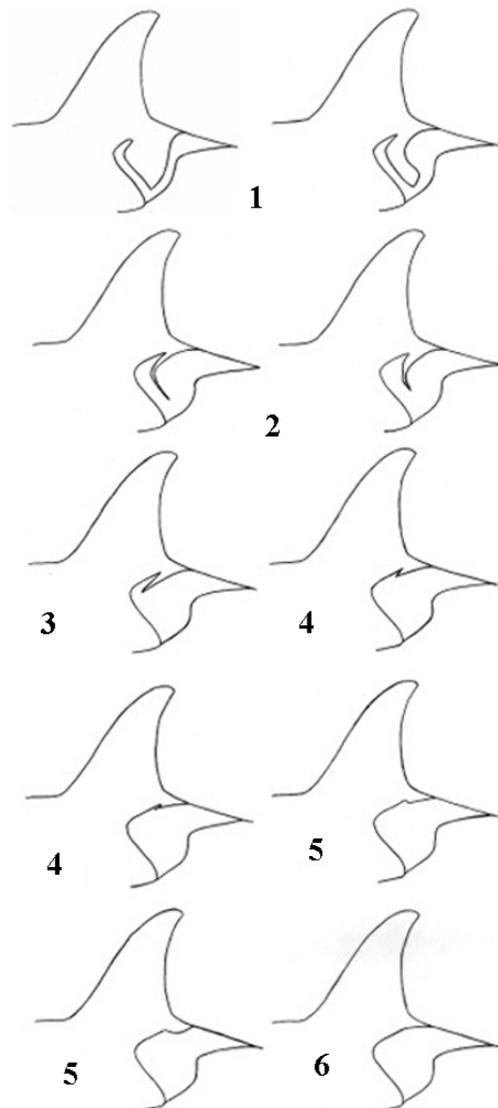


Fig. 1. Six types of saddle patch shape used for the analysis of phenotypic differences.

Analysis of social structure was performed using SOCPROG 2.4 software (Whitehead, 2006), which conducts the statistical analysis of social associations. For every animal the simple ratio association index was calculated as $AI_{AB} = N_{AB} / (N_{AB} + N_A + N_B)$ where AI_{AB} is the association index between individual A and individual B, N_{AB} is the number of times individual A was in association with individual B (see association definition in main text); N_A is the number of times individual A was not associating with individual B; and N_B is the number of times individual B was without individual A. Then, using linkage-average cluster analysis, dendrogram was constructed that graphically illustrates level of association among animals and between units.

The initial acoustic analysis was performed in AviSoft SASLab Pro software. After looking through recordings and selecting good-quality calls, we split stereotyped calls into structural elements – syllables, separated by pauses or frequency shifts. Contours of syllables were extracted using MATLAB software. Extracted contours were compared using a dynamic time-warping algorithm. The distances obtained from this method were used to build a dendrogram.

Biopsy skin samples were used to analyse feeding ecology and genetic structure. To study feeding habits, we conducted the analysis of the ratio of stable isotope ^{15}N , which increases with trophic level of the animal. Samples were dried and ground to a powder to ensure homogenization. Aliquots (0.5 to 0.6 mg) of a homogenized sample were sealed in tin capsules and then analyzed using a Thermo-Finnegan Delta V Plus Mass Spectrometer coupled to a Flash 1112 Elemental Analyzer at the A.N. Severtsov Institute of Ecology and Evolution RAS (Moscow, Russia). Stable isotope ratios were reported as per mille (‰) using delta notation determined from the equation: $\delta X = ((R_{\text{sample}}/R_{\text{standard}}) - 1) \times 1000$, where X is ^{15}N or ^{13}C and R is the corresponding ratio of $^{15}\text{N}/^{14}\text{N}$ or $^{13}\text{C}/^{12}\text{C}$. Standard reference materials were atmospheric nitrogen gas and carbon from Pee Dee Belemnite (VPDB).

For the analysis of genetic population structure, we analyzed the allele frequency of nine nuclear DNA microsatellite loci in skin samples from Avacha Gulf, Karaginsky Gulf, waters of Bering Island (Commander Islands) and from the western Sea of Okhotsk (total 67 samples).

Results

All transient killer whales in our study had closed saddle patches. Resident killer whales had ~70% closed (type 6) and ~30% open and semi-open saddle patches (type 1 – 3%, type 2 – 5%, type 3 – 2%, type 4 – 6%, type 5 – 13%).

The analysis of social associations shows that associations between whales were non-random. Most of animals had maximal SRI greater than 0.5. Using dendrogram and data from direct observations we defined 36 units of killer whales. From comparing some morphology features such as shape of dorsal fin and saddle patch, and behavioral

features, members of six units were identified as transient killer whales. Statistical analysis showed that transient units did not have any associations with resident units. Transient killer whales are clearly isolated from resident killer whales in this area; they do not form mixed groups or aggregations with each other. Structure of groups was also different between resident and transient killer whales: residents travelled in large stable groups, while transients occurred in small and less stable groups. Solitary males were observed in transient, but not in resident killer whales.

Behaviour observations of foraging showed that resident killer whales were feeding on salmon and cod. We did not observe any hunting behavior of transient killer whales, but our colleagues working at fur seal rookery at the same island reported that they regularly observed killer whales hunting on fur seals. We compared our photo-catalogs and found that some whales identified by us as transient were observed feeding on fur seals, but no matches between our resident killer whales and their seal-hunters were found. Moreover, we regularly observed fur seals fearlessly following the groups of resident killer whales (fig. 2).



Fig. 2. Fur seals following the group of resident killer whales.

Besides, we had reports from our colleagues from Avacha Gulf and western Okhotsk Sea about killer whales feeding on minke whale and bearded seals. Biopsy samples of these whales showed that they belonged to transient ecotype both by genetic and stable isotope analysis (see below).

Acoustic monitoring showed that transient killer whales were silent most of the time, while resident killer whales were much more vocal. Few recordings of transient sounds showed clear differences from calls recorded from resident killer whales. The syllables of resident

killer whales fell into three discrete clusters: syllables of the high-frequency component, syllables of the low-frequency component and short transitional syllables. Transient killer whales showed a different picture. Also, the substantial differences between ecotypes were found in frequency parameters of syllables of stereotyped calls. Syllables of both high- and low-frequency components of transient killer whales were lower, than those of resident killer whales. In transient killer whales, high-frequency syllables had a frequency of around 5 kHz. Syllables of low-frequency component of transient killer whales were in the frequency range of up to 1.5-2 kHz. In the same time, high-frequency syllables of resident killer whales mostly were in the range from 5 to 11 kHz, while low-frequency syllables were in the range of up to 4 kHz.

Genetic analysis demonstrated clear differences between residents and transients. Cluster analysis (program Structure 2.3.3) showed that the samples were more likely to be divided into two groups, rather than the other number of groups (1, 3-5) (fig. 3). Killer whales from different ecotypes were the most different from each other. At the same time some heterogeneity among animals of transient type can indicate their belonging to different reproductive groups. Average allelic diversity of killer whales from transient ecotype was much higher than those values for resident killer whales, which may also indicate that transient animals belong to several communities, but clearly distinguish them from resident whales. A certain degree of reproductive isolation of individuals between different ecotypes was also confirmed by further analysis of data from nine loci. Differences of resident killer whales from transient whales were statistically significant and relatively high. However, we found no significant differences in allele frequencies between groups of killer whales within each ecotype. We conclude that killer whales from different ecotypes – resident and transient - found in the study areas, represent different reproductive groups.

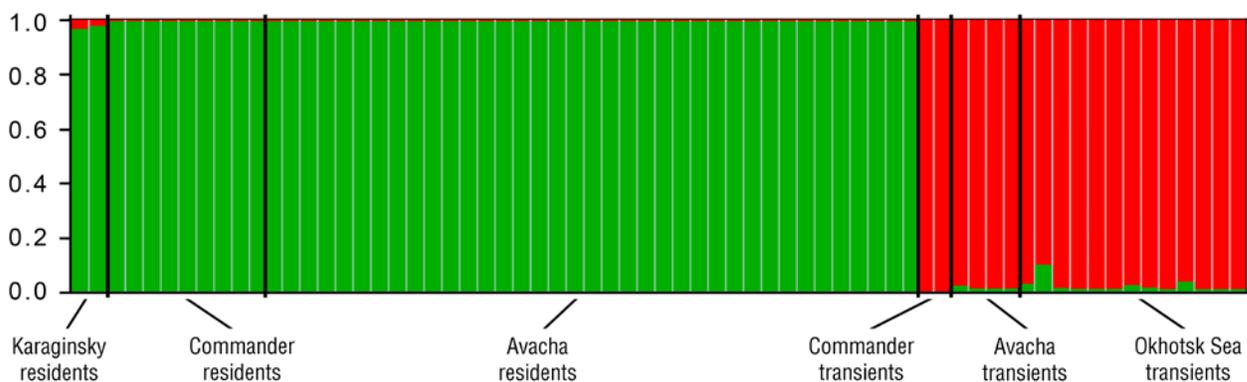


Fig. 3. Probability of belonging of samples to one of two reproductive clusters. Each column represents one sample, colours reflect the probability of belonging to one of the clusters.

Analysis of stable isotopes showed that ratio of ^{15}N (and, therefore, the trophic level) was lower in resident than in transient killer whales (median \pm SE: residents 13.79 \pm 0.76, transients 16.76 \pm 0.94) (fig. 4). Difference in ratio of ^{15}N between resident and transient killer whales was statistically significant (Mann-Whitney test, $p < 0.001$). This confirms our observations that resident killer whales feed on fish, while transient killer whales hunt on marine mammals.

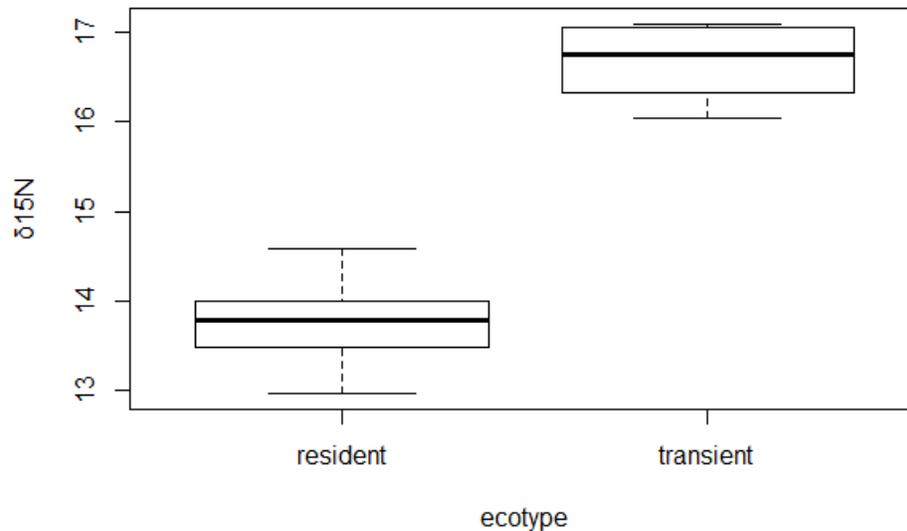


Fig. 4. Ratio of ^{15}N in skin samples of resident and transient killer whales.

Conclusions

Our data clearly shows that resident and transient killer whales meet the definition of species in biological species concept, since they represent sympatric reproductively isolated populations with stable ecological, morphological and behavioural differences.

Communicatory and popularization activities

To disseminate the results among the public, we had written a popular scientific article for Russian journal "Priroda" ("Nature"), it will be published in May 2013. We gave talks to the local people in Nikolskoye village (Commander Islands). We informed the public about our progress on [our website](#) and on [our Facebook page](#) which currently has more than 2000 "likes".

To disseminate the results among the scientists, I gave many talks at conferences, workshops and meetings. I participated in the 26th Conference of the European Cetacean Society in Dublin, March 2012 with the presentation "Diversity of monophonic and biphonic calls in mammal-eating and fish-eating killer whales of the North Pacific".

I organised killer whale workshop in frames of VII International Conference «Marine mammals of the Holarctic» in Suzdal (Russia) in September 2012 (see the workshop description [here](#), workshop proceedings [here](#)). Twenty experts from three countries (Russia, Canada, USA) attended the workshop. All of them agreed that our data clearly demonstrated the existence of two reproductively isolated sympatric ecotypes in the waters of the Russian Far East.

On 4 December 2012 I gave a talk "New data on the population structure of killer whales of the Russian Far East: suggestions for population management" at the meeting of the Russian Marine Mammal Council. The report emphasized the need for separate assessment of abundance of killer whales from fish-eating (resident) and mammal-eating (transient) populations and claimed that the estimate of 2.5-3 thousand killer whales in Okhotsk Sea, calculated by VNIRO (All-Russian Research Institute of Fisheries and Oceanography), violated generally accepted standards of marine mammal accounts and did not consider the separation of fish-eating and mammal-eating populations of killer whales. The reaction of the Council to the report was generally positive, but faced strong opposition from Dr. A.I. Boltnev (VNIRO). Dr. Boltnev said that fish-eating and mammal-eating specialization of killer whales is the result of individual strategies, and he claimed he did not believe that killer whales of different food specialization belong to different populations (see video (turn on English subtitles) [here](#)). He did not bother to take into account our references to multiple publications of foreign scientists, and questioned the results of our own genetic analysis. The debate on this issue has been blocked by the Chairman of the Council academician A.V. Yablokov, due to time constraints. As a result, the Council did not accept our suggestion for separate assessment of fish-eating and mammal-eating killer whales.

On 22 January, the Russian Marine Mammal Council held its meeting where live-capturing of killer whales, walrus and beluga whales was discussed. Again, I presented the recent scientific data on the population structure of killer whales and noted that these data were ignored when assessing the total allowable catches for the species. Furthermore, in my report, I showed that the abundance estimates used to calculate the total allowable catches of orcas in the Sea of Okhotsk were not reliable, and the level of reproduction of killer whales populations, which is needed to calculate the total allowable catches, was never assessed by fisheries bodies. After the discussion, the Council finally decided to recommend to change the procedure for evaluating the total allowable catches and fishing regulations in accordance with the scientific data. The Council decided to form the Workgroup on Killer Whales and put me in charge of it. After this, I persuaded the Council to write the official letters to Russian Federal Fisheries Agency and to Russian Research Institute of Fisheries and Oceanography (organizations in charge of capture quotas)

claiming that the numbers of resident and transient killer whales must be estimated separately. These letters are currently under discussion in these organizations.

I have also contacted the Commission on the New Edition of the Russian Red Book and suggested including transient killer whales into the New Edition of the Russian Red Book, which is now being discussed by the committee.

Now we prepare a scientific paper for the peer-reviewed Russian journal based on the results of our project to inform the broader scientific audience about the problem.

Web-site

We continue to put on [our website](#) and on [our Facebook page](#) information about killer whales in the wild and the problem of their capturing. We were the first to attract the public attention to the information about the young transient orca captured in August 2012 in western Okhotsk Sea. We pioneered the campaign to free this orca (called "Narnia" by internet orca-lovers, see "Free Narnia" campaign) and provided the information about her to the public, which the capturing company desired to keep in secret. We have a good feedback on our forum, and we hope that our activity will contribute to the rise of concern of Russian web-users towards marine mammals.